Remarks

Applicant and Applicant's representatives thank the Examiner and SPE Prouty for the helpful suggestions provided during the March 27, 2002, Examiner interview.

Reconsideration of this Application is respectfully requested. Upon entry of the foregoing amendment, claims 1, 3, 5-10, 13, 16, 17, 19, 26, 28, 29, 34-38, 40-44 are pending in the application, with claims 1, 37 and 38 being the independent claims. Claims 1, 5, 6, 37-38 and 40 were amended as discussed during the Examiner interview. Support for the amendment can be found, *inter alia*, at page 4, lines 3-14; at page 6, lines 1-5; at page 18, line 26 to page 19, line 25; and at page 21, line 21 to page 22, line 14. Applicants reserve the right to pursue the subject matter of the unamended claims in continuing applications. Claims 4 and 39 were canceled without prejudice or disclaimer. Claims 36 and 44 were amended to correct a typographical error. Support for this amendment can be found, *inter alia*, at page 7, lines 5-18.

These changes are believed to introduce no new matter, and their entry is respectfully requested. Based on the above amendment Applicants respectfully request that the Examiner reconsider all outstanding objections and rejections and that they be withdrawn.

Conclusion

All of the stated grounds of objection and rejection have been properly accommodated or rendered moot. Applicants therefore respectfully request that the

Examiner reconsider all presently outstanding objections and rejections and that they be withdrawn. Applicants believe that the present application is in condition for allowance. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Prompt and favorable consideration of this Amendment is respectfully requested.

Respectfully submitted,

STERNE, KESSLER, GOLDSTEIN & FOX P.L.L.C.

Gaby L. Longsworth

Gaby L. Longsworth, Ph.D.
Agent for Applicant
Registration No. 47,756

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1100 New York Avenue, N.W.
Suite 600
Washington, D.C. 20005-3934
(202) 371-2600

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Version with markings to show changes made

In the Claims:

Claims 4 and 39 were canceled without prejudice or disclaimer.

The following claims 1, 5, 6, 36-38, 40 and 44 were substituted for pending claims 1, 5, 6, 36-38, 40 and 44:

1. (Once amended) A *Thermotoga maritima* (*Tma*) DNA polymerase mutant which is modified at least two ways selected from the group consisting of:

(a) a mutation in the 3'-5' exonuclease domain of said polymerase to reduce or eliminate the 3'→5' exonuclease activity of the polymerase;

(b) a mutation in the 5'-3' exonuclease domain of said polymerase to reduce or eliminate the 5'→3' exonuclease activity of the polymerase; and

(c) a mutation in the O-helix of said polymerase to reduce or eliminate discriminatory behavior against a dideoxynucleotide.

5. (Once amended) The DNA polymerase of claim [4] 1, wherein said O-helix is defined as RXXXKXXXFXXXYX (SEQ ID NO:1), wherein X is any amino acid.

6. (Once amended) The *Tma* DNA polymerase as claimed in claim [5] 1, wherein said mutation in the O-helix is a $Phe^{730}\rightarrow Tyr^{730}$ substitution.

36. (Once amended) A method of sequencing a DNA molecule, comprising:

(a) hybridizing a primer to a first DNA molecule;

(b) contacting said DNA molecule of step (a) with deoxyribonucleoside triphosphates, the DNA polymerase of claim 1, and a terminator nucleotide;

(c) incubating the mixture of step (b) under conditions sufficient to synthesize a random population of DNA molecules complementary to said first DNA molecule, wherein said synthesized DNA molecules are shorter in length than said first DNA molecule and wherein said synthesized DNA molecules comprise a terminator nucleotide at their [5´] 3' termini; and

(d) separating said synthesized DNA molecules by size so that at least a part of the nucleotide sequence of said first DNA molecule can be determined.

37. (Once amended) An isolated DNA molecule encoding a *Thermotoga maritima* (*Tma*) DNA polymerase mutant which is modified at least two ways selected from the group consisting of:

(a) a mutation in the 3'-5' exonuclease domain of said polymerase to reduce or eliminate the 3' 5' exonuclease activity of the polymerase;

(b) a mutation in the 5'-3' exonuclease domain of said polymerase to reduce or eliminate the 5' 3' exonuclease activity of the polymerase; and

(c) a mutation in the O-helix of said polymerase to reduce or eliminate discriminatory behavior against a dideoxynucleotide.

38. (Once amended) A mutant *Tma* DNA polymerase having a mutation in the O-helix, resulting in said DNA polymerase becoming non-discriminating against

dideoxynucleotides, or a fragment of said mutant DNA polymerase said fragment having polymerase activity.

40. (Once amended) The mutant *Tma* DNA polymerase of claim [39] 38, wherein said O-helix is defined as RXXXKXXXFXXXYX, wherein X is any amino acid.

44. (Once amended) A method of sequencing a DNA molecule, comprising:

(a) hybridizing a primer to a first DNA molecule;

(b) contacting said DNA molecule of step (a) with dexyribonucleoside triphosphates, a DNA polymerase of claim 38, and a terminator nucleotide;

(c) incubating the mixture of step (b) under conditions sufficient to synthesize a random population of DNA molecules complementary to said first DNA molecule, wherein said synthesized DNA molecules are shorter in length than said first DNA molecule and wherein said synthesized DNA molecules comprise a terminator nucleotide at their [5'] 3' termini; and

(d) separating said synthesized DNA molecules by size so that at least a part of the nucleotide sequence of said first DNA molecule can be determined.